Seventh Quarterly Report

Date of Report: *March 14, 20010*

Contract Number: DTPH56-08-T-000014

Prepared for: DOT-PHMSA

Project Title: Effect of Concentration and Temperature of Ethanol in Fuel Blends on Microbial and Stress Corrosion Cracking of High-Strength Steels

Prepared by: Colorado School of Mines and National Institute of Standards and Technology

Contact Information: Dr. *David Olson, dolson@mines.edu, Dr. Brajendra Mishra, bmishra@mines.edu, Dr. John Spear, jspear@mines.edu, Dr. Shaily Bhola sbhola@mines.edu, Luke Jain ljain@mines.edu, Chase Williamson chawillia@gmail.com, Dr. Tom Siewert, siewert@boulder.nist.gov, Dave McColskey, mccolske@boulder.gov, Timothy Weeks, timothy.weeks@nist.gov, Mark Richards mark.richards@nist.gov, Jeffrey Sowards jeffrey.sowards@nist.gov*

For quarterly period ending: *March 14, 2010*

Technical Status

Technical efforts for this quarter have included field sampling from ethanol infrastructure, microbiological analysis of field samples, inoculation of steel coupons for evaluation of surface corrosion, inoculation of specimens for cyclic loading tests, microbial isolation experiments, preparation for 454 pyrosequencing, development of long term field testing of steel coupons, assembly and calibration of the MSBT, development of FCGR testing equipment, and presentation of a technical paper at an international conference.

Field Assessment and Microbiological Characterization

FGE containment tanks that capture ethanol spillage resulting from normal operation at fueling terminals have experienced corrosion problems. In some instances, tanks have been reported to smell like vinegar (acetic acid). As microbes are known to produce acetic acid while using ethanol as a substrate, they may be active in the ethanol contact-water tanks. Additional tank bottoms samples from ethanol contact-water tanks have been acquired from fueling terminals. Experiments designed to isolate microbes that metabolize iron and gasoline from these samples have been initiated (Figure 1).

Fig. 1: Tube containing E10, water, and iron filings inoculated with media from an ethanol contact water tank.

DNA has been extracted from a number of samples collected from ethanol industry infrastructure, and these samples will be analyzed with 454 pyrosequencing (preparation in progress). 454 pyrosequencing is a next-generation sequencing technology that will produce a large amount of DNA sequence data that will provide greater insight into the types of microbes present in environments containing fuel grade ethanol.

Arrangements have been made to collect additional samples from ethanol industry infrastructure, which will allow for a more thorough understanding of the types of microbes that are associated with these environments.

Corrosion Evaluation of Long Term Field Coupons

Microbial diversity and microbiologically influenced corrosion (MIC) is being evaluated in ethanol fuel environments. While water in fuel grade ethanol (FGE) tanks is less likely to be concentrated in the bottom of the tank as it will likely dissolve into the concentrated ethanol, water in ethanol fuel tanks with lower concentrations of ethanol such as E-10 will be more likely to maintain a free water layer beneath the fuel. This water layer could be able to harbor microbial growth and promote MIC in the same way that is encountered in diesel and kerosene storage systems.

The proposed testing will evaluate corrosion of welded ASTM A36 carbon structural steel field coupons immersed in FGE and ethanol fuel blends (EFBs) in atmospheric fuel storage tanks. Stresses in the specimen will be developed by the welding process only. Observations from the tests will be used to determine the types of corrosion attack that occur in tank bottoms containing ethanol fuels. The data will be used to measure the corrosion performance of welded ASTM A36 carbon steel in ethanol fuels.

The welding process and parameters are intended to approximate those employed in the base weld of a vertical atmospheric AST. Residual stress from the weldment in combination with surface flaws may increase susceptibility to initiation of surface corrosion and SCC. Artificial pits mimicking surface flaws will act as stress concentrators, provide for localized regions near the fuel-metal interface where the fuel may have different electrochemical properties then in the bulk, and serve as sheltered areas that may be more favorable to microbial growth.

Testing Objectives include:

1) Evaluate corrosion resistance of the entire weldment (weld, fusion line, HAZ, base metal).
2) Characterize any corrosion in the specific zones of the weld (weld, fusion line, HAZ, base metal).
3) Evaluate the effect of stress raisers in the weldment.
4) Evaluate the effect of stress raisers away from the weldment.
5) Characterize microbial diversity associated with corroded regions of the coupon

Rectangular coupons will be saw-cut from 0.25-inch ASTM A36 welded carbon steel plate. The field coupons will be cut into rectangular coupons 3 inches by 4 inches. The coupons will retain the thickness of the original 0.25-inch ASTM A36 plate.

The welding process will be bead-on-plate. Weld ends will be discarded. Coupons will be removed from across the weld. No loading (dynamic or static) will be applied to the welded coupons.

Common welding processes used for tank fabrication include welding with 6010 stick, E7024 Jet Rod, FCAW, and SAW. Specimens containing welds formed by each of these techniques will be submersed in each testing environment.

A relatively simple bead-on-plate weld was selected for the field coupons as it still incorporates residual stress into the weldment but allows for a simple coupon shape and weldment geometry.

Several pits will be machined into the coupon to emulate surface flaws. The pits will be placed in the weld metal, heat affected zone (HAZ), base metal adjacent to the HAZ, and base metal away from the HAZ. These will act as stress concentrators and provide for localized regions near the fuel-metal interface where the fuel may have different electrochemical properties then that in the bulk. These may also serve as sheltered areas that may be more favorable to microbial growth.

The surface of the coupon will be left in the as-fabricated condition as the effects of surface conditions are being evaluated on initiation of surface corrosion.

A microstructural characterization will be performed for the weld metal, weld interface, and HAZ. Chemical composition will be determined for the weld metal and base metal. Mechanical properties will be characterized for the material.

Prior to exposure, the coupons will be cleaned and evaluated for the presence of cracks and weld surface defects. Any defects present will be documented prior to immersion. All specimens will also be weighed to the nearest 0.1 mg. The total surface area will be determined to an accuracy of +/- 1 pct.

The specimen rack will also be fabricated out of 0.25-inch ASTM A-36 carbon structural steel. While the rack may corrode, the overall integrity of the system is not expected to compromise during the testing duration. The specimen rack will consist of two plates in parallel with each other as well as the specimens that will be fastened between them. Teflon spacers will offset and insulate the specimens from the specimen rack. The two plates and specimen will be bolted together. Insulating sleeves will separate the specimens from the bolts. Double nuts will be used to prevent loosening during the testing duration. All fasteners will be fabricated from stainless steel to discourage corrosion during exposure. Two additional holes will be machined into one plate of the rack to provide a location to secure a nylon rope that will be used to lower and raise the rack once it is in the tank.

Four specimens will be spaced 0.125 inches apart on each test rack. Each specimen will be stamped with an identification number.

The field specimens will be immersed in the stagnant bottom of atmospheric fuel storage tanks for the duration of the test. The coupons will experience conditions (degree of aeration, fuel composition, contaminate levels, solid settlements) consistent with those existing in the tank bottoms.

The specimen rack will be lowered into the fuel storage tank by the nylon rope. The rack will be positioned so that it is resting on the bottom of the tank for the duration of the test. The rack will be constructed so that the coupons will not contact any tank surfaces. The rope will be secured on the inside of the manway. Testing duration will be two months. The duration may be modified to be made more convenient to operations and maintenance schedules.

The coupons will be inspected for:

1) The presence of surface corrosion, including cracking and pitting, over a given interval
2) The location of the surface attack (weld metal, weld interface, HAZ, base metal)
3) The microstructure of the cracked area and the depth and intensity of cracking

Photographs will be taken of the specimen after removal from the test environment. Samples of any corrosion products or films will be recovered for evaluation of chemical composition.

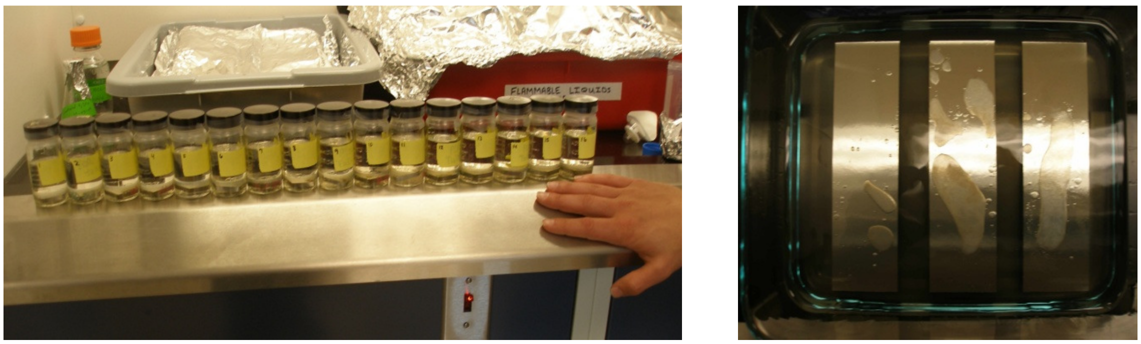
Specimens will be cleaned according to ASTM G1. Photographs will be taken of the cleaned surface.

Low-magnification (1 – 20X) examination will be used to evaluate surface attack such as stress cracking or pitting. Higher magnification (100X) will be used to evaluate the presence of smaller cracks; however, the as fabricated surface condition may limit the resolution of smaller cracks.

Pitting will be evaluated according to ASTM G46. The number, size, distribution, general shape, and uniformity of the pits will be noted. The maximum and minimum pit depths will be measured with a calibrated microscope or by use of a depth gage. Penetration damage will be reported in millimeters (nearest 0.01mm).

Laboratory cultivation efforts

Cultivation and corrosion testing efforts have included experiments designed to evaluate MIC of steel coupons in water and E10 environments inoculated with a variety of environmental microbes (Figure 2). Pitting and surface damage of these coupons will be monitored with time. Specimens for cyclic loading tests have also been inoculated with environmental microbes to determine the potential corrosion effects of microbes on loaded coupons in water and E10 environments (Figure 2).



(a) (b)

Fig. 2: (a) steel coupons in water and E10 inoculated to evaluate MIC (b) steel specimens inoculated for cyclic loading tests that will determine the effects of MIC on loaded specimens in water and E10 environments

Previous electrochemical experiments have shown an interesting corrosion phenomenon on steels exposed to ethanol containing low amounts of water (0-10% by volume). Experiments are underway to determine if microbes may play a role in corrosion under these conditions. Tubes containing ethanol with low concentrations of water (0-10%) have been inoculated with a variety of environmental microbes to evaluate the potential for microbes to survive and/or effect corrosion in these environments.

Multi-Specimen Bend Testing

Five specimens of type A36 steel were machined to the same tolerances as proposed test coupons for four point bend testing. Strain gages were applied to the top and bottom faces of the specimens, where the specimen top is to be loaded in tension and the specimen bottom is to be loaded in compression. The gage configuration was placed such that strain measurements were made along the center of the stress fields as indicated in Figure 3. The figure shows the positioning of the strain gages installed on the specimens in part (a) and the loading-configuration (b) indicating top and bottom strain gages for measuring tensile and compressive stresses, respectively. Figure 3 (c) shows the location of the strain channels for the following results. The specimens were used to determine required forces and displacement distances required to achieve equivalent strains during four point bending in solutions. The procedure is reported as follows.

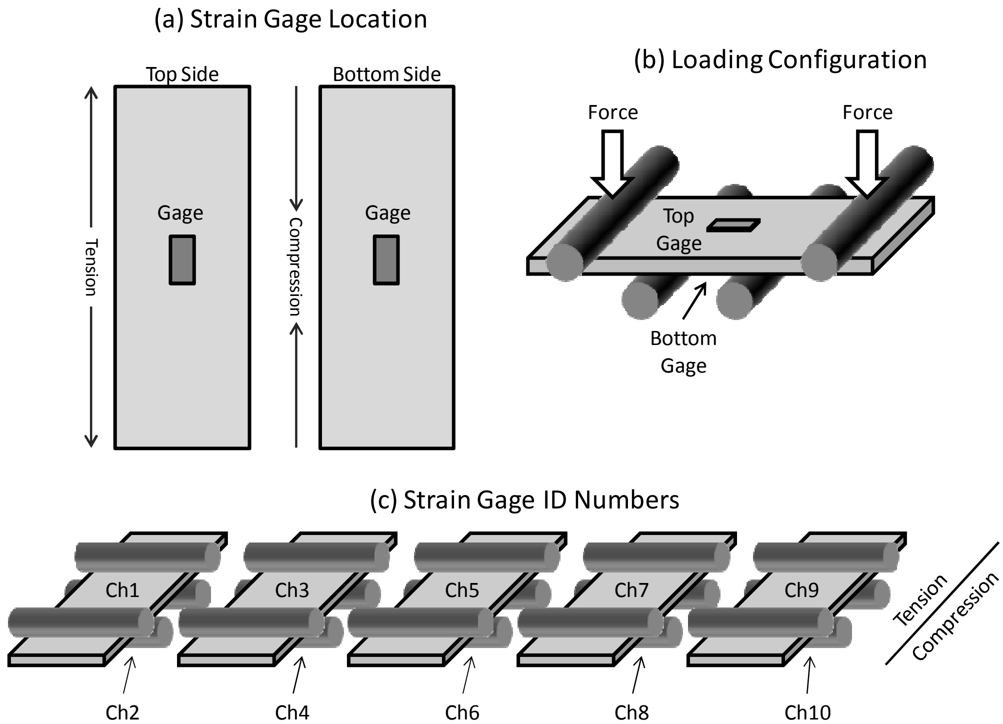
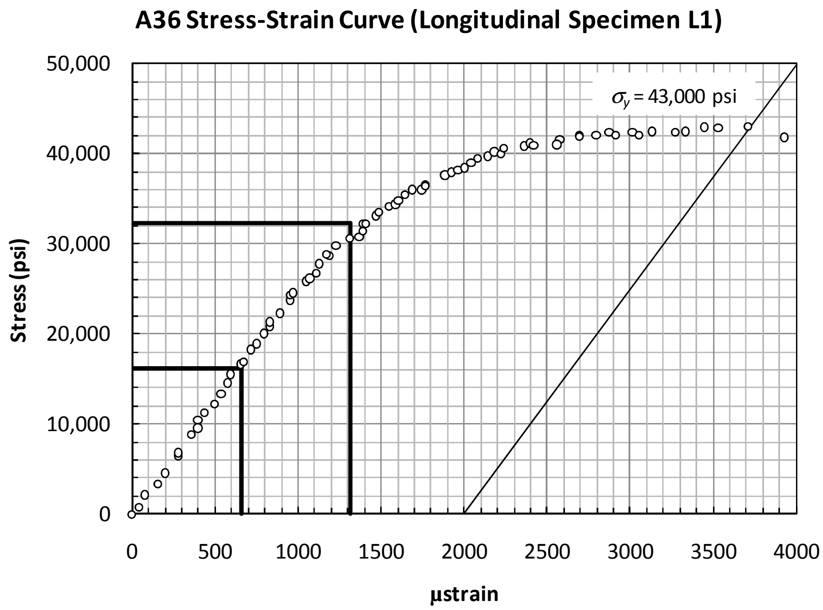


Fig. 3: Schematic illustration of (a) strain gage positioning, (b) loading configuration used for calibration, and (c) sample layout.

Maximum stress levels during four point bend testing have been selected to be 75% of the yield stress. The min stress was selected to be half the maximum stress ($R = 0.5$), approximately 37.5% of the yield stress. Actual stress strain data was measured for the A36 steel in the longitudinal and transverse directions as reported in the last quarterly report. The transverse data showed slightly higher yield and ultimate tensile strength levels, as well as slightly lower elongation until failure. Therefore, for the bend testing, the lower values of stress determined for the longitudinal tests were used for a conservative estimate of yield strength of the bend test specimens. Yield stress was found to be approximately 43,000 psi as shown in the stress strain curve of Figure 4. Minimum and maximum stress levels for four point bend testing were determined from the data as shown in the enlarged stress-strain curve of the elastic region. The minimum (37.5%) and maximum (75%) stress levels are indicated on the Figure and were calculated to be 16,125 and 32,250 psi, respectively. The strain levels required to achieve these stress levels were found to be 660 and 1,320 micro strain respectively based on the stress-strain curve. Therefore, these strain values were used to determine required load levels and displacements for the four point bending mechanical tests.



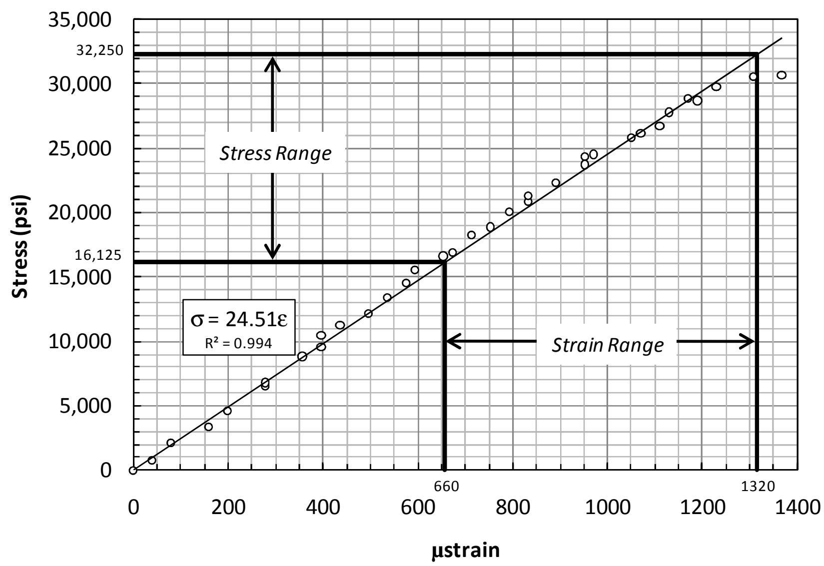


Fig. 4: Tensile stress-strain curve of A36 steel (top) used for four point bend testing calibration specimens, and (bottom) enlarged version of linear elastic region showing suggested ranges of stress and strain levels selected for testing.

The five gauged specimens were placed in the loading fixture and simultaneously loaded. Strain levels were recorded as a function of the crosshead displacement. Displacement control was used since that will be the method of control during actual testing in fuel blends. Strain is shown as a function of displacement in Figure 5(a). Note that there was significant variation (200 µstrain) in recorded strain levels between the specimens. The loading rollers were adjusted at each specimen using the threaded assemblies to achieve similar strain levels at each specimen for a fixed load or displacement. This was done to ensure that when loading specimens during testing, minimum and maximum stress levels will be approximately the same for all five specimens. After several iterative adjustments were made to achieve uniform loading at each specimen, strain vs displacement was recorded again as shown in Figure 5(b). Deviations in strain were reduced to approximately ± 20 µstrain in the range where testing will be performed. This strain variation correlates to maximum stress variations of approximately 500 psi among the five samples.

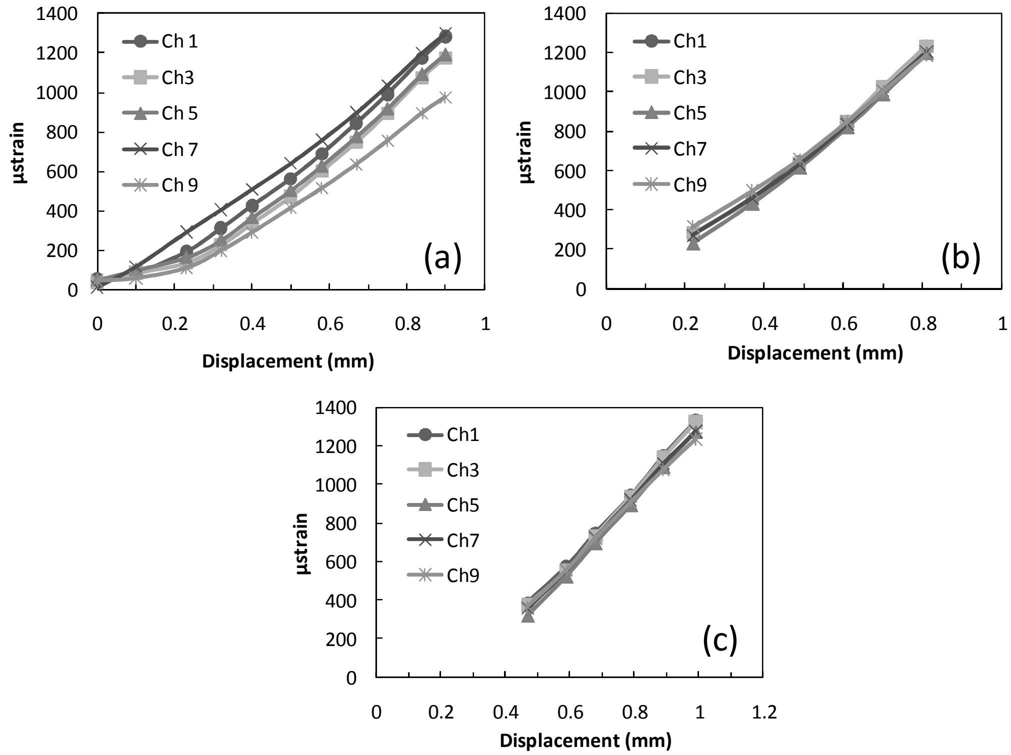


Fig. 5: Strain-displacement curves for the four point bend tests (a) prior to adjustment, (b) after adjustments were made to compensate for strain variations, and (c) after 50+ cycles of loading at 0.1 Hz.

After adjustments were made to achieve uniform loading, the specimens were subjected to the cyclic loading procedure that will be used for actual testing. Using displacement control, the specimens were loaded from approximately 600 to 1300 µstrain at a frequency of 0.1 Hz. A plot of crosshead displacement as a function of time is shown in Figure 6. Note that the test control is capable of performing the test as intended. The specimens were cycled 50+cycles. The strain-displacement curves were evaluated again after the cyclic loading as shown in Figure 5(c). Note that the strain levels are still within approximately 40 µstrain. This strain variation correlates to maximum stress variations of approximately 980 psi among the five specimens.

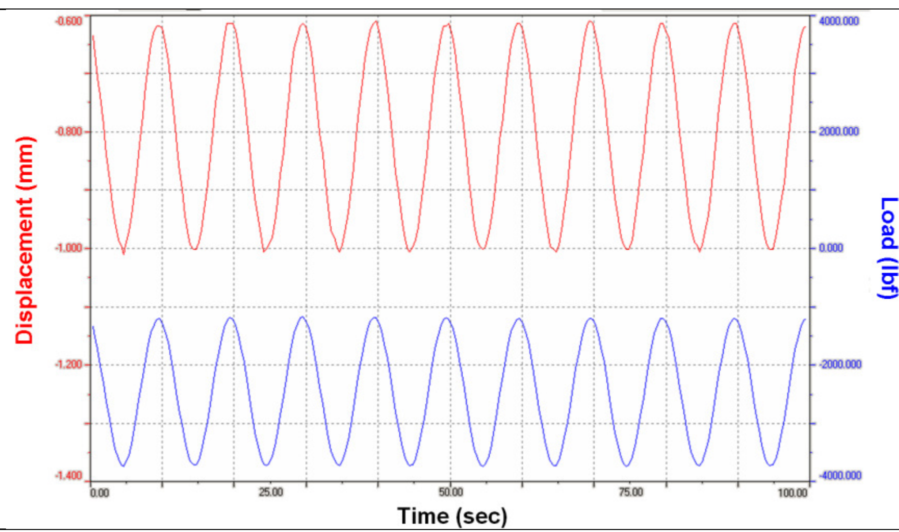


Fig. 6: Displacement and loading as a function of time for the cyclic loading of four point bend tests.

The strain data collected after cyclic testing, shown in Figure 5(c), was used to calculate stresses at the location of the strain gages. Stress was calculated with the equation shown in the lower part of Figure 4, assuming that the testing was within the elastic regime. Resulting stress-strain curves at the location of the strain gages are shown in Figure 5. Note that tensile and compressive stresses show excellent agreement on each plot as shown in Figure 7.

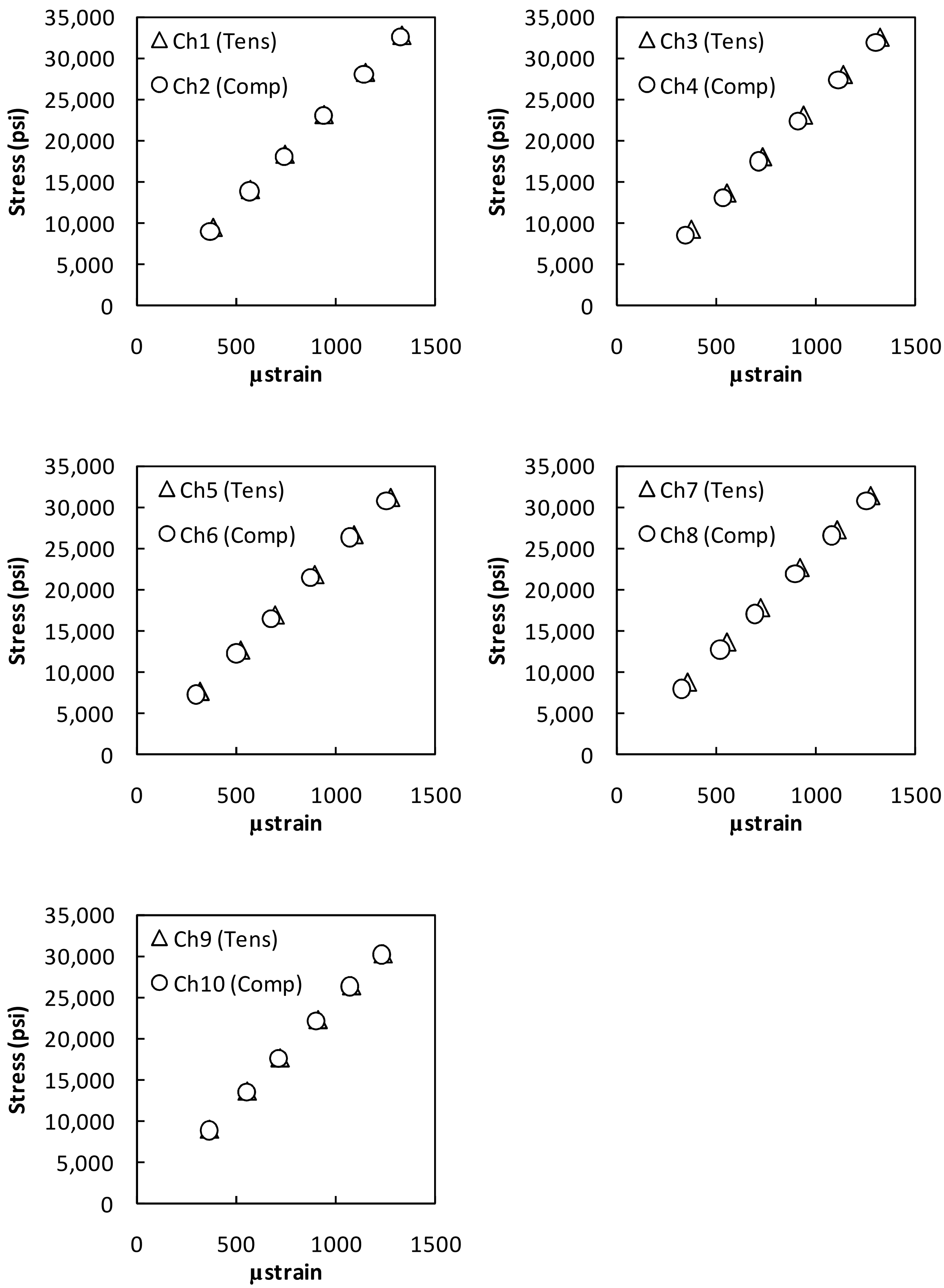


Fig. 7: Stress-strain curves for four point bend test specimens.

Safety

A detailed standard operating procedure (SOP) has been drafted for mechanical testing in mixed fuel solutions at the NIST facility. This SOP is currently under review by the NIST safety office and must be approved before testing in the various fuel solutions can commence. Fume extraction systems are currently being designed to contain and remove fuel vapors from the laboratory. This system must also be completed before testing. Lastly, a safety review will be performed after the SOP and fume extraction system are both in place.

Conference Paper Submission

The paper "Microbiological and Electrochemical Evaluation of Corrosion and Microbiologically Influenced Corrosion of Steel in Ethanol Fuel Environments" was presented at CORROSION 2010 Conference and Expo (March 14 – 18, 2010, San Antonio, TX, U.S.A.). during the Biofuel Corrosion Issues Symposium March 16, 2010: 2:00 PM-6:00 pm).

Results and Conclusions:

- Microbial growth and corrosion experiments have continued to determine the ability of microbes to affect corrosion in high-ethanol environments.
- Analyses of samples collected from fueling terminals handling fuel grade ethanol have begun. DNA sequencing will provide insight into the types of microbes present at these fueling terminals.
- Methods to sterilize fuels/liquids for microbial cultivation, extract DNA from corroded specimens, and visualize microbes in corrosion product samples are being investigated.
- The isolation of manganese-oxidizing spores from Bacillus sp. cultures is underway and preliminary experiments have begun to determine if these spores cause corrosion of iron/steel.
- Contacts were made with additional researchers evaluating microbial activity in ethanol fuel environments.
- Construction of all components for the MSBF has been completed.
- Calibration of the MSBF has been completed
- A gas purging system for the MSBF has been designed and components have been purchased.
- Construction of a ventilation system for the MSBF and CT testing has been completed.
- Sampling supplies have been shipped to JIPs for field sample collection.
- Mechanical test frames were installed in the NIST facility for corrosion evaluation of ethanol fuels. The environmental test chambers for bend testing have been assembled onto the load test frame.
- NIST received shipment of the new controller for Fatigue Crack Growth studies.
- A hazard assessment of the NIST facility for corrosion evaluation of ethanol fuels was completed and an SOP has been drafted.
- The mechanical testing lab at NIST in Boulder, CO is being equipped with an *in situ* ventilation system to vent any ethanol/fuel vapor safely out of the lab per safety requirements.
- Pre-testing incubation of specimens for MSBF testing has begun.
- Steel coupons for long-term field-testing of steel coupons were designed.
- Sites suitable for field coupon placement were located and selected using JIP participating.
- JIPs were contacted with regard to field-testing. A JIP member has offered to fabricate the coupons as a cost-share.
- The paper on electrochemical analysis of mild carbon steel has been submitted to an archival journal.
- A presentation highlighting a technical paper summarizing experimental work performed for the project thus far was presented fat NACE International 2010 Corrosion Conference and Expo.

Issues, Problems or Challenges

Gaining access to infrastructure for sampling purposes

Plans for Future Activity:

- Conduct further thin-film and growth experiments to determine the potential for microbes to survive in high ethanol environments and the potential for spore-forming microbes to effect corrosion
- Continue to analyze 16S rRNA gene data as well as conduct cultivation experiments to support field assessment
- Begin testing with the MSBF
- Develop and refine a MIC/ethanol review paper
- Continue experiments to determine the ability of microbes, including spores, to affect corrosion in high-ethanol environments.
- Pursue sampling opportunities and collect samples from EFB infrastructure.
- Calibrate the new controller for Fatigue Crack Growth studies.
- Construct fume containment and extraction hood for the bend tests to ensure fuel vapors are properly vented from test facility.
- Approve an SOP for mechanical testing in mixed fuel environments.
- Begin mechanical loading of four point bend and CT specimens.
- Continue efforts to collaborate with other scientists working on microbial contamination in ethanol fuel blends.
- Fabricate coupons for long-term field-testing.
- Field coupons will be placed.